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Investigation and evaluation of ultrasound reactor for reduction of fungi from sewage*

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Abstract: The objective of the investigation was to study the application of ultrasound reactor technology (USRT) as a disinfectant for reduction of fungi from sewage effluent. Fungi are carbon heterotrophs that require preformed organic compounds as carbon sources. USRT is an attractive means to improve water quality because of the system simplicity and no production of toxic by-products. An ultrasound reactor produces strong cavitation in aqueous solution causing shock waves and reactive free radicals by the violent collapse of the cavitation bubble. These effects should contribute to the physical disruption of microbial structures and inactivation of organisms. There was significant reduction in fungal growth, with decreased fungal growth with increasing USRT. In this study, ultrasound irradiation at a frequency of 42 kHz was used to expose suspensions of fungi to evaluate the disinfection efficacy of the ultrasound reactor. Also, this study showed that in this system more than 99% reduction of sewage fungi was achieved after 60 min.

Key words: Ultrasound reactor, Sewage, Fungi, Cavitation, Frequency
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INTRODUCTION

Fungi importance

It has been estimated that there are approximately 1.5 million species of fungi and among these approximately 400 species have so far been found to cause disease in humans and animals. Fungi including yeasts and filamentous species or molds are ubiquitously distributed, achlorophyllous, heterotrophic organisms with organized nuclei and usually with rigid walls. Fortunately normal healthy individuals rarely suffer from serious fungal diseases; it is immunocompromised individuals that are at risk of fatal fungal infections. Water used in hospitals to wash burns, flush eyes, mix solutions and other uses where it will come in contact with injured or damaged tissues should be sterile. Any water used for people with compromised immune systems must also be sterile since pathogenic fungi are a serious risk for such people (Anon, 2000; Deinega, 1986).

Fungi and their spores are ubiquitous. Fungi are present in, and have been recovered from, diverse, remote, and extreme aquatic habitats including lakes, ponds, rivers, streams, estuaries, marine environments, wastewaters, sludge, rural and urban storm water runoff, well waters, acid mine drainage and aquatic sediments (APHA, 1989).

Historical background

The basis for the present-day generation of ultrasound was established as far back as 1880 with the discovery of the piezoelectric effect by the Curies. Most modern ultrasonic devices rely on transducers (energy converters), which are composed of piezoelectric materials. Such materials respond to the application of an electrical potential across opposite faces with a small change in dimension. This is the

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inverse of the piezoelectric effect. If the potential is alternated at high frequencies, the crystal converts the electrical energy to mechanical vibration (sound) energy—like a loudspeaker. At sufficiently high alternating potential high frequency sound (ultrasound) will be generated. When more powerful ultrasound at a lower frequency is applied to a system, it is possible to produce chemical changes as a result of acoustically generated cavitation. Cavitation as a phenomenon was first identified and reported in 1895 by John Thornycroft and Sidney Barnaby. Since l945, an increasing understanding of the phenomenon of cavitation has developed coupled with significant developments in electronic circuitry and transducers (i.e. devices which convert electrical to mechanical signals and vice versa). As a result of this there has been a rapid expansion in the application of power ultrasound to chemical processes, a subject that has become known as "Sonochemistry" (Gelate *et al*., 2000; Suslick, 1994).

Sound theory

Ultrasound irradiation is a sound with a pitch so high that the human ear cannot hear it. Frequencies above 18 kHz are usually considered to be ultrasonic. The frequencies used for ultrasonic cleaning range from 20000 kHz to over 100000 kHz. The most commonly used frequencies for industrial cleaning are those between 20 and 50 kHz. Frequencies above 50 kHz are more commonly used for high precision cleaning, removal of small particles and delicate parts (Gelate *et al*., 2000; Suslick and Price, 1999).

Ultrasound has wavelengths between successive compression waves measuring roughly 10 to 10^{-3} cm. These are not comparable to molecular dimensions (Fig.1). Because of this mismatch, the chemical effects of ultrasound cannot result from a direct interaction of sound with molecular species (Suslick, 1994; Gong and Hart, 1998).

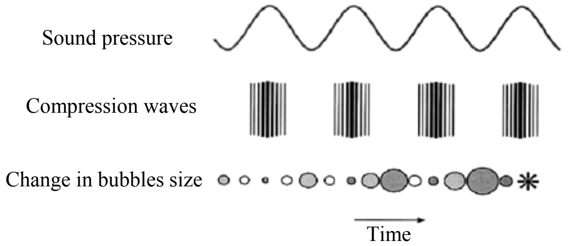

Fig.1 The compression and expansion cycle of ultrasound

Bubble cavitation

Ultrasound reactor technology (USRT) in a liquid leads to the acoustic cavitation phenomenon, such as the formation, growth, and collapse of bubbles, accompanied by the generation of local high temperature, pressure, and reactive radical species. The basis for ultrasound irradiation applications is that acoustic cavitation can effect a number of mechanical, acoustic, chemical and biological changes in a liquid (Lauterborn and Ohl, 1997; Laborde, 1998). In elastic media such as air and most solids, there is a continuous transition as a sound wave is transmitted. In non-elastic media such as water and most liquids, there is continuous transition as long as the amplitude or "loudness" of the sound is relatively low. As amplitude is increased, however, the magnitude of the negative pressure in the areas of rarefaction eventually becomes sufficient to cause the liquid to fracture because of the negative pressure, causing a phenomenon known as cavitation. Cavitation "bubbles" are created at sites of rarefaction as the liquid fractures or tears because of the negative pressure of the sound wave in the liquid. As the wave fronts pass, the cavitation "bubbles" oscillate under the influence of positive pressure, eventually growing to an unstable size. Finally, the violent collapse of the cavitation "bubbles" results in implosions, which cause shock waves to be radiated from the sites of the collapse. The collapse and implosion of myriad cavitation "bubbles" throughout an ultrasonically activated liquid result in the effect commonly associated with ultrasound (Hua and Hoffmann, 1997; Kalumuk, 2003; Neppiras, 1980).

Types of acoustic cavitation

There are two types of acoustic cavitation: transient and stable (or controlled). Transient cavities exist for a few cycles, and are followed by a rapid and violent collapse, or implosion, that produces very high local temperatures. Ultrasonic cleaning frequencies, typically between 20 and 350 kHz, transform low-energy/density sound waves into high-energy/density collapsing bubbles, producing transient acoustic cavitation. Transient acoustic cavitation can cause damaging surface erosion in more sensitive substrates. Megasonic frequencies, 700 to 1000 kHz, produce stable acoustic cavitation bubbles, which have less time to grow, and are smaller, resulting in a less vigorous collapse than in transient cavitation. The implosion associated with these smaller, gas-filled bubbles is less likely to produce surface damage. Thus, megasonic cavitation is better suited for sensitive substrate surfaces (Joyce *et al*., 2002; Laborde, 1998; Suslick and Crum, 1997; Neppiras, 1980).

Ultrasound disinfection importance

Recently, disinfection by USRT has been studied and is another important disinfection method in practice. The possible mechanisms by which cells are rendered inviable during ultrasound irradiation include free-radical attack, including hydroxyl radical attack, and physical disruption of cell membranes (Scherba *et al*., 1991; Phull *et al*., 1997). Once the cell membrane is sheared, chemical oxidants can enter the cell and attack internal structures, or vital structures can be released from the cell, and degraded in solution. Furthermore, ultrasound irradiation can facilitate the disagglomeration of microorganisms and thus, increase the efficiency of other chemical disinfectants (Hua and Thompson, 2000; Scherba *et al*., 1991; Petrier, 1992; Phull *et al*., 1997).

EXPERIMENTAL METHODS

Experimental apparatus

Controlled laboratory experiments have established the ability of this system to destroy sewage fungi. The ultrasound reactor (Fig.2) was applied to samples using a laboratory-cleaning bath with the characteristics shown in Table 1.

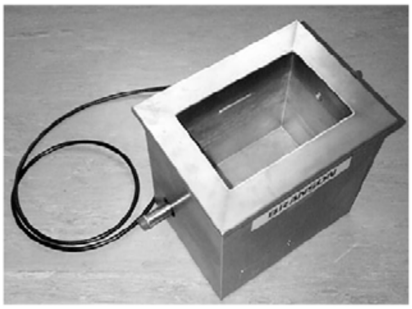

Fig.2 Laboratory ultrasound reactor for reduction of fungi

Table 1 Characteristics of ultrasound reactor

Parameters	Characteristics	Parameters	Characteristics
Input	220~230 V	Capacity	1.5 L
Output	70 W	Reactor type	Basin
Power	155 W	Manufacturer	K.G. Co., Germany
Frequency	42 kHz		

Biological experiments

Aqueous solution was sonicated in a temperature-controlled system. Sonication was performed at laboratory temperature. The pH was 6.5~7.0 without chemicals addition. Before ultrasound, the number of fungi in solution was determined by colony counting. Also, after ultrasound the number of surviving fungi colonies was determined by colony counting. Results are as colony-forming units (CFU) per 100 ml original sample. In lab-scale experiments ultrasound system operated at 42 kHz was used to sonicate sewage samples. The disinfection times for experiments were 10, 15, 20, 25, 30, 35, 40, 45, 50, 55 and 60 min. For each trial, sample of polluted water fungi was exposed for each of the durations. The number of trials per exposure level was variable. All the analyses were performed according to the procedures outlined in standard methods (APHA, 1989).

This research will provide basic information on the fundamentals of USRT as a novel disinfection technology for sewage fungi destruction. This study carried out in the laboratory of Medical Sciences/ University of Tehran in 2005.

RESULTS

The results of disinfection during sonicating 500 ml fungi suspension at eight different samples (200, 1000, 2000, 3500, 5500, 6500, 10000 and 17000 CFU/ml) are shown in Fig.3. The number of fungi decreases with increasing disinfection time. The results showed that increasing the disinfection time has a significant effect on fungi reduction. Also, there is no significant reduction of fungi in less than 15 min exposure time to 42 kHz but considerable levels of reduction can be expected after longer periods (99.92%). Table 2 summarizes the average results of these studies. The data presented in Table 2 are from experiments conducted on eight samples. All cases are seen to produce rapid reduction in fungi cells.

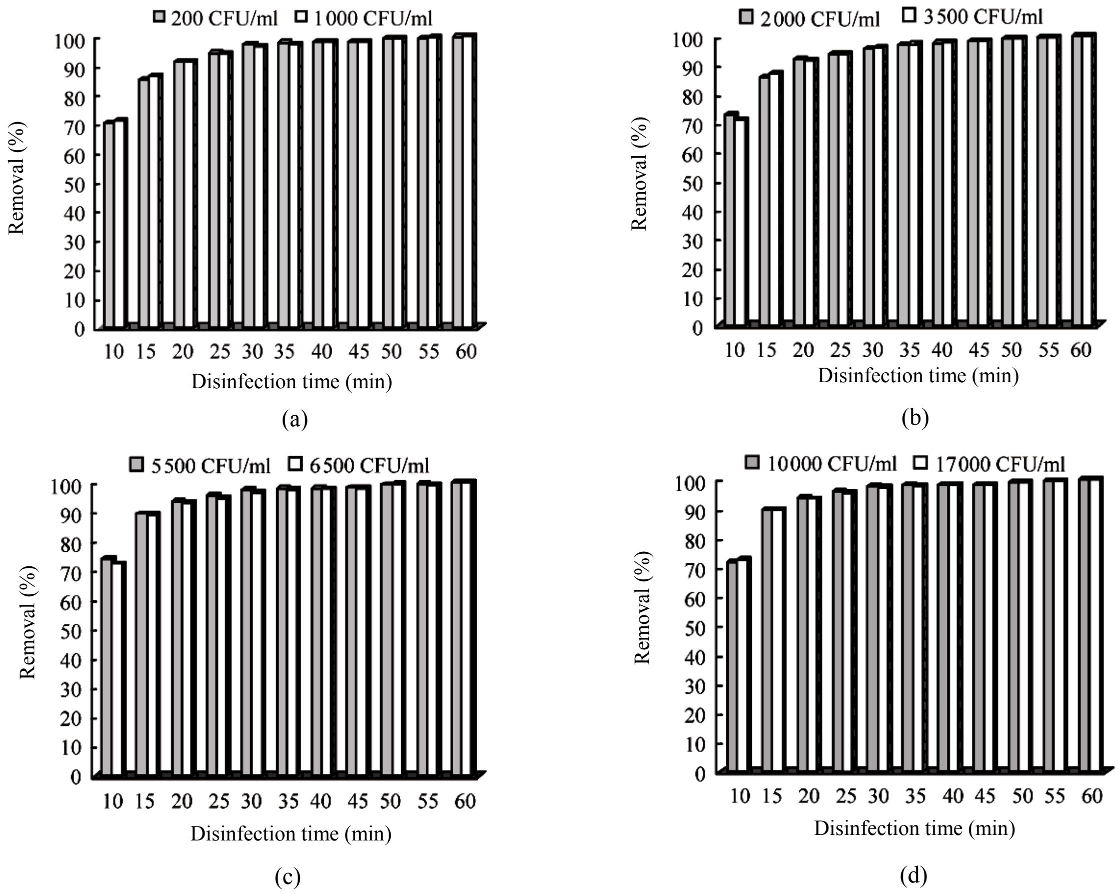

Fig.3 Comparison of plate count monitoring at different samples. (a) 200 and 1000 CFU/ml; (b) 2000 and 3500 CFU/ml; (c) 5500 and 6500 CFU/ml; (d) 10000 and 17000 CFU/ml

Table 2 Average of removal percentage of fungi

Disinfection time (min)	Average removal (%)	Disinfection time (min)	Average removal (%)
10	71.85	40	97.88
15	88.09	45	98.17
20	92.41	50	99.21
25	94.59	55	99.43
30	96.76	60	99.92
35	97.52		

Scherba *et al.*(1991) suggested that USRT at a frequency of 26 kHz is capable to some degree of inactivating fungi cells. Experiments at 42 kHz can be seen to be more effective than operation at less than this frequency.

Tsukamoto *et al.*(2004) suggested that in a squeeze-film-type sonicator, more than 90% inactivation of fungi was achieved for 60 min. In this experiment, sonolytic inactivation of fungi cells was investigated using a horn-type sonicator at 27.5 kHz frequency. The results of the fundamental investigation included effect of USRT power, cell numbers, and flow rate on the inactivation of the fungi cells using a horn-type sonicator and a squeeze-film-type sonicator. Inactivation by USRT was fastest at the lowest initial cell numbers.

Different authors showed that the reduction of organisms is mainly due to cavitation. During this process younger cells are more affected because they are much more sensitive (Everett, 1978; Phull *et al.*, 1997; Scherba *et al.*, 1991).

DISCUSSION

In this study, the major objective of this work was to study, investigate and apply USRT as a novel disinfection technology for reduction of sewage fungi. The reduction of fungi was carried out by an ultrasound reactor. The results for the removal of sewage fungi using ultrasound reactor are reported in Fig.3.

According to Table 2 the highest and lowest fungi reduction after disinfection by USRT was 99.92% and 71.85%. On the other hand, the results showed decreased growth of sewage fungi with increasing disinfection time at 42 kHz frequency.

Also, USRT was used to expose samples of sewage fungi to evaluate the germicidal efficacy of USRT. There was a significant effect of time to kill fungi, with percent killed increasing with increased duration of sonication.

CONCLUSION

USRT substantially improves the effectiveness of removing sewage fungi through the effects of acoustic cavitation in water. Transient cavitation and stable cavitation need to be considered in order to gain an understanding of what cavitation like activity might be responsible for the reduction of sewage fungi. In propagated ultrasound reactor, transient cavitation process occurs more easily at lower ultrasound frequency. As a result, USRT is suitable for disinfection of sewage fungi. For effective reduction of fungi using USRT alone it is almost certain that USRT would need to be applied in combination with another common disinfection technologies used in water treatment including ultraviolet irradiation, ozone or chlorination.

USRT is a very small unit that easily can be installed at any place in a treatment plant. Quality USRT can replace sand filters that usually serve as a step to remove suspended solids prior to disinfection. There is scientific and economic potential in the development of combined disinfection processes. In order to definitely damage sewage fungi walls higher USRT energy input is necessary. Also, combination with another disinfectants applications is useful.

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